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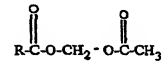
(54) Novel acetyloxymethyl esters and methods for using same

(57) Novel acetyloxymethyl esters having the formula (1):



(1)

wherein R is as defined in the description, are disclosed. Use of these compounds in treating an illness, including cancer, hemological disorders and inherited metabolic disorders, and treating or ameliorating other conditions using these compounds are also disclosed. The compounds are effective in the inhibition of histone deacetylase.

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Description

FIELD OF THE INVENTION

[0001] The present invention relates to novel acetyloxymethyl esters. The invention further relates to use of those acetyloxymethyl esters in the treatment of cancer and other proliferative diseases, hemoglobinopathies and inherited metabolic disorders and to treat or ameliorate various other illnesses and conditions, such as by hematopoietic stimulation. The present invention also relates to methods for using the disclosed compounds in the inhibition of histone deacetylase.

BACKGROUND INFORMATION

[0002] Histones are unique proteins in the nucleus of a cell. DNA is wound around a complex of histones to form nucleosomes. As such, histones are an integral structural element of the chromatin material. The histones complexed with the DNA are susceptible to a range of chemical modifications, one of which is acetylation, and the reverse of which is deacetylation. Acetylation of histone protein is believed to facilitate transcription of the DNA, thereby enhancing correlating with gene expression. Histone deacetylase is believed to reverse the process that represses gene expression. Histone dynamics are regulated by two enzymes--histone acetyl transferase and histone deacetylase.

[0003] Hyperacetylation due to inhibition of histone deacetylation, and the resulting expression of a latent gene, have been observed or proposed to occur in numerous inherited metabolic diseases and in cancer. The inhibition of histone deacetylase is believed to activate an otherwise dormant fetal gene, which serves as a redundant or back-up gene. Pharmacological inhibition of histone deacetylase, therefore, is believed to induce the expression of repressor genes in cancer tissue, inhibit the expression of tumor-promoting genes, and induce the expression of the redundant or back-up gene in patients suffering from various metabolic and hematological diseases. Thus, inhibition of histone deacetylase is proposed to slow the growth of neoplastic cells and/or reverse the deficient process of various metabolic and hematological diseases. Inhibition of histone deacetylase is also believed to play a role in antiprotozoal activity.

[0004] Trichostatin is the most potent inhibitor of histone deacetylase observed so far, but due to various drawbacks, such as availability of the material, has not been pursued.

[0005] Butyric acid is a natural product that has been known for several decades to be an effective differentiating and antiproliferative agent in a wide spectra of neoplastic cells *in vitro*. For example, butyric acid has been reported to induce cellular and biochemical changes in cancer cells, to induce apoptosis, and to increase the expression of transfected DNA, although the mechanism of action of butyric acid is unknown. Increased histone acetylation following treatment with butyric acid has been correlated with changes in transcriptional activity and at differentiated states of cells. Butyric acid and its salts, however, have shown low potency in both *in vitro* assays and clinical trials, and thus require large doses to achieve even minimal therapeutic effects. This can lead to fluid overload and mild alkalosis.

[0006] The present invention is directed to acetyloxymethyl ester compounds, and methods for using the same, that have also been found to inhibit histone deacetylase. The present compounds show significantly greater activity than butyric acid or its salts. That acetyloxymethyl esters such as those of the present invention have the ability to inhibit histone deacetylase has been previously unreported in the art.

SUMMARY OF THE INVENTION

[0007] The present invention relates to novel acetyloxymethyl esters as described below. The compounds are useful in the inhibition of histone deacetylase.

[0008] The present invention therefore further relates to methods of treating a patient for an illness, particularly wherein the illness is one in which histone deacetylase inhibition would be beneficial. Examples include cancer, hemoglobinopathies and inherited metabolic disorders. Other illnesses and conditions that can be treated according to the present invention are discussed herein. In the case of histone deacetylase inhibition, the present compounds are believed to function by chelating the zinc ion at the active site of histone deacetylase; the inventor does not wish to be bound by this mechanism, however.

[0009] It is therefore an aspect of the invention to provide novel acetyloxymethyl esters.

[0010] Another aspect of the invention provides methods for treating a patient using acetyloxymethyl esters.

Use of the present acetyloxymethyl esters for the manufacture of a medicament for treating an illness in a patient, particularly where the illness is one in which histone deacetylase inhibition would be beneficial, is also an aspect of the invention.

[0011] A method for inhibiting histone deacetylase in a patient is also an aspect of the present invention.

Use of the present acetyloxymethyl esters for the manufacture of a medicament for inhibiting histone deacetylase in a patient is also an aspect of the present invention.

[0012] These and other aspects of the invention will be apparent upon reviewing the attached specification and appended claims.

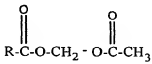
BRIEF DESCRIPTION OF THE FIGURES

[0013]

Figure 1 shows a reaction scheme for phenylpropioloyloxymethyl acetate, prepared according to example 1.
Figure 2 shows a reaction scheme for S-benzylthioglycoloyloxymethyl acetate, prepared according to Example 2.
Figure 3 shows a reaction scheme for 3-(phenylthio)acryloyloxymethyl acetate, prepared according to Example 3.
Figure 4 shows a reaction scheme for 4-cyclohexanebutyroyloxymethyl acetate, prepared according to Example 4.
Figure 5 shows a reaction scheme for 6-phenyl-3,5-hexadienoyloxymethyl acetate, prepared according to Example 5.
Figure 6 shows a reaction scheme for 5-phenyl-2,4-pentadienoyloxymethyl acetate, prepared according to Example 6.
Figure 7 shows a reaction scheme for cinnamoyloxymethyl acetate, prepared according to Example 7.

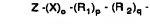
DETAILED DESCRIPTION OF THE INVENTION

[0014] The present invention is directed to acetyloxymethyl esters having the general formula (1):



(1)

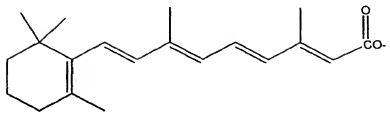
wherein R is a substituted or unsubstituted C_2-C_7 alkenyl, a substituted or unsubstituted C_2-C_7 alkynyl, a cis or trans retinoyl group, or has the general formula (2):



(2)

wherein Z is selected from the group consisting of hydrogen and substituted or unsubstituted aryl, heteroaryl, cycloalkyl having the formula C_nH_{2n-1}, and alkoxy;
wherein n is 3 or greater;
wherein X is S, O, C=O or CH_2;
wherein R_1 is S, O, CH=CH or C≡C;
wherein R_2 is CH_2, CH=CH or C≡C; and
wherein o, p and q are the same or different are each between 0 and 7, but when o is zero and R_1 or R_2 is CH=CH or C≡C, Z is not hydrogen or alkoxy.

[0015] A retinoyl group will be understood as being derived from retinoic acid and having general formula (3):



(3)

[0016] Preferred embodiments of formula 1 include the following:

phenylpropioloyloxymethyl acetate (wherein R is C_6H_5-C≡C-);
S-benzylthioglycoloyloxymethyl acetate (wherein R is C_6H_5-CH_2-S-CH_2-);
3-(phenylthio)acryloyloxymethyl acetate (wherein R is C_6H_5-S-CH=CH-);
4-cyclohexanebutyroyloxymethyl acetate (wherein R is C_6H_{11}-$(CH_2)_3$-);
cinnamoyloxymethyl acetate (wherein R is C_6H_5-CH=CH-);
6-phenyl-3,5-hexadienoyloxymethyl acetate (wherein R is C_6H_5-$(CH=CH)_2$-CH_2-);
and 5-phenyl-2,4-pentadienoyloxymethyl acetate (wherein R is C_6H_5-$(CH=CH)_2$-).

[0017] As used herein, "aryl" includes any stable 5- to 14-membered monocyclic, bicyclic or tricyclic ring, containing at least one aromatic carbon ring, for example, phenyl, naphthyl, indenyl, tetrahydronaphthyl (tetralinyl) and the like. "Stable" as used herein refers to compounds that are sufficiently robust to survive isolation to a useful degree of purity from a reaction mixture. As noted above, one or more substituents on the aryl group are optional, and when present, the substitutents can be the same or different and can be halogen, alkyl, alkoxy, hydroxy, cyano, amino, nitro, carbonyl or halogenated hydrocarbon groups.

[0018] As used herein, the term "heteroaryl" includes any stable 5- to 14-membered monocyclic, bicyclic or tricyclic aromatic heterocyclic ring comprising carbon atoms and from 1 to 5 heteroatoms selected from the group consisting of N, O and S, wherein the nitrogen may optionally be quaternized; the term includes any bicyclic or tricyclic group in which a heteroaryl ring is fused to the other ring(s). The heteroaryl ring may be attached to its pendant group at any heteroatom or carbon atom that results in a stable structure. As with the aryl group, the presence of substitution on the heteroaryl group is optional; if present, the one or more substituents can be on a carbon atom or heteroatom so long as the resulting compound is stable and all the valencies of the atom have been satisfied. The one or more substituents on the heteroaryl group can be the same or different and are the same as those substituents listed above for the aryl groups; also included are alkylammonium salts when the substituent is an alkyl group attached to the nitrogen atom of the heteroaryl ring. These quarternized ammonium salts include halides, hydrohalides, sulfates, methanesulfates, toluenesulfates, nitrates, phosphates, maleates, acetates, lactates or any other pharmaceutically acceptable salt. Examples of heteroaryl groups include, but are not limited to, furanyl, thienyl, pyrrolyl, pyrazolyl, imidazolyl, tetrazolyl, benzofuranyl, benzothienyl, indolyl, indolenyl, quinolinyl, isoquinolinyl, benzimidazolyl, thiazole, thioxane, benzothiozole and benzothiazin.

[0019] As discussed above, either of the Z groups can be a cycloalkyl group having the general formula C_nH_{2n-1} wherein n is 3 or greater; any stable cycloalkyl group having this general formula is therefore within the scope of the invention. Typically, "n" will not be higher than about 12. Again, the cycloalkyl can be unsubstituted or substituted with one or more of the substituents listed above. Similarly, Z can be an alkoxy group which is unsubstituted or substituted with one or more of the same substituents. "Alkoxy" will be understood by those skilled in the art as referring to an alkyl group having at least one oxygen substituent represented by R-O, wherein R is an alkyl group having between about 1 and 5 carbons.

[0020] The term "alkyl" is used herein to refer to branched or straight chain saturated aliphatic hydrocarbon groups. Unless indicated otherwise, the alkyl groups typically have 15 carbons or less, e.g. 1-6 carbons.

[0021] Pharmaceutically acceptable salts of any of the above compounds are also within the scope of the invention. As used herein, "pharmaceutically acceptable salts" refer to derivatives of the disclosed compounds that are modified

by making acid or base salts. Examples include, but are not limited to, mineral or organic acid salts of basic residues such as amines, alkali or organic base salts of acidic residues such as carboxylic acids, and the like. Pharmaceutically acceptable salts include, but are not limited to, hydrohalides, sulfates, methanesulfates, toluenesulfonates, nitrates, phosphates, maleates, acetates, lactates and the like.

[0022] Pharmaceutically-acceptable salts of the compounds of the present invention can be prepared by reacting the free acid or base forms of these compounds with a stoichiometric or greater amount of the appropriate base or acid in water or in an organic solvent, or in a mixture of the two; generally, nonaqueous media like ether, ethyl acetate, ethanol, isopropanol, or acetonitrile are preferred. The salts of the invention can also be prepared by ion exchange, for example. Lists of suitable salts are found in *Remington's Pharmaceutical Sciences*, 18th ed., Mack Publishing Company, Easton, PA (1990).

[0023] The present invention is also directed to methods of treating illnesses in which proliferation of neoplastic or diseased cells occurs, or illnesses in which inhibition of histone deacetylase would be desired; as discussed above, inhibition of histone deacetylase results in the enhancement of gene expression that slows the growth of neoplastic cells and reverses the deficient process of various metabolic and hematological diseases. Accordingly, the present invention is also directed to the use of the compounds in the manufacture of a medicament for treating illnesses in which proliferation of neoplastic or diseased cells occurs, or illnesses in which inhibition of histone deacetylase would be desired; as discussed above, inhibition of histone deacetylase results in the enhancement of gene expression that slows the growth of neoplastic cells and reverses the deficient process of various metabolic and hematological diseases. It will be understood that the present invention encompasses the treatment of various illnesses, as that term is defined herein, regardless of whether the treatment is through histone deacetylase inhibition, through another mechamism, or through a variety of mechanisms. The present compounds have a plasma half-life of sufficient length to effect a therapeutic benefit without requiring excessive doses, or doses that are toxic to a patient.

[0024] More specifically, the present invention is further directed to a method for treating an illness in a patient comprising administering to that patient an effective amount of a compound having general formula (1):

(1)

wherein R is substituted or unsubstituted C_2-C_{10} alkenyl, substituted or unsubstituted C_2-C_{10} alkynyl, a cis or trans retinoyl group, or has the general formula (2):

$$Z\text{-}(X)_o\text{-}(R_1)_p\text{-}(R_2)_q\text{-} \qquad (2)$$

wherein Z is selected from the group consisting of hydrogen and substituted or unsubstituted aryl, heteroaryl, cycloalkyl having the formula C_nH_{2n-1}, and alkoxy;

wherein n is 3 or greater;

wherein X is S, O, C=O or CH_2;

wherein R_1 is S, O, CH=CH or C≡C;

wherein R_2 is CH_2, CH=CH or C≡C; and

wherein o, p and q are the same or different are each between 0 and 10, but when o is zero and R_1 or R_2 is CH=CH or C≡C, Z is not hydrogen or alkoxy.

[0025] Illnesses treatable according to the present invention include, but are not limited to, various cancers, hematological diseases, and inherited metabolic diseases. Cancer includes, but is not limited to, leukemias, such as acute promyelocytic leukemia, acute myeloid leukemia, and acute myelomonocytic leukemia; other myelodysplastic syndromes; multiple myeloma such as breast carcinomas, cervical cancers, melanomas, colon cancers, nasopharyngeal carcinoma, non-Hodgkins lymphoma (NHL), Kaposi's sarcoma, ovarian cancers, pancreatic cancers, hepatocarcinomas, prostate cancers, squamous carcinomas, other dermatologic malignancies, teratocarcinomas, T-cell lymphomas, lung tumors, gliomas, neuroblastomas, peripheral neuroectodermal tumors, rhabdomyosarcomas, and prostate tumors and other solid tumors. Hematological diseases or hemoglobinopathies within the scope of the present invention in-

clude, but are not limited to, thalassemias, sickle cell anemias, infectious anemias, aplastic anemias, hypoplastic and hypoproliferative anemias, sideroblastic anemias, myelophthisic anemias, antibody-mediated anemias, anemias due to chronic diseases and enzyme-deficiencies, and anemias due to blood loss, radiation therapy and chemotherapy. Inherited metabolic diseases include, but are not limited to, Duschenne's muscular dystrophy, cystic fibrosis, and adrenoleukodystrophy. Thus, the term "illness" as used herein encompasses at least all of these things.

[0026] The term "illness" as used herein also encompasses various conditions such as cutaneous ulcers and gastrointestinal disorders. The cutaneous ulcers which can be treated in accordance with the current methods include leg and decubitus ulcers, stasis ulcers, diabetic ulcers and atherosclerotic ulcers. Gastrointestinal disorders treatable by the present methods include colitis, inflammatory bowel disease, Crohn's disease and ulcerative colitis. The term illness also refers to wounds such as abrasions, incisions, and burns.

[0027] "Illness" also encompasses treatment, prevention, or amelioration of virus-associated tumors including, but not limited to, EBV-associated malignancy, Kaposi's sarcoma, AIDS-related lymphoma, hepatitis B-associated malignancy or hepatitis C-associated malignancy. EBV-associated malignancy include, but are not limited to, nasopharyngeal carcinoma and non-Hodgkins' lymphoma. The present compounds can be administered in conjunction with a therapeutically effective amount of an antiviral agent such as ganciclovir, acyclovir and famciclovir. Protozoan infections are also included within "illness" and include, for example, malaria, cryptosporidiosis, trypanosomiasis, Eimeria sp., Plasmodium sp., toxoplasmosis, and coccidiosis.

[0028] In another embodiment of this invention, "illness" refers to alopecia, or hair loss. Alopecia is a common condition that results from diverse causes. In particular, alopecia frequently occurs in cancer patients who are treated with chemotherapeutic drugs and/or irradiation. Such agents damage hair follicles which contain mitotically active hair-producing cells. Such damage may cause abnormally slow growth of the hair or may lead to frank loss. Thus, the present invention further relates to methods for protecting against injury to hair follicles in a patient by administering one or more of the present compounds to the patient.

[0029] "Patient" refers to members of the animal kingdom, including but not limited to humans. Preferably, the methods of the present invention are applied to a patient suffering from any of the illnesses listed above.

[0030] The compounds of the invention can be administered by any conventional means available for use in conjunction with pharmaceuticals, either as individual therapeutic agents or in combination with other therapeutic agents known in the art for the illness being treated. The compounds can be administered alone, but are generally administered with a pharmaceutically acceptable carrier selected on the basis of the chosen route of administration and standard pharmaceutical practice. The compositions of the invention may be adapted for oral, parenteral, topical, transdermal, transmucosal, rectal or intranasal administration, and may be in unit dosage form; the term "parenteral" as used herein includes subcutaneous, intravenous, intramuscular, or intrasternal injection or infusion techniques. Methods for preparing the present compositions for use by a patient are well known to those skilled in the pharmaceutical arts; formulations can include one or more fillers or preservatives in addition to the active ingredient and carrier.

[0031] "Pharmaceutically acceptable carrier" includes any and all solvents, dispersion media, coatings, antibacterial and antifungal agents, isotonic and absorption delaying agents, and the like. The use of such media and agents for pharmaceutically active substances is well known in the art. Use of any of these media or agents is contemplated with the compounds of the present invention, absent compatibility problems with the active compound.

[0032] It is especially advantageous to formulate compositions in dosage unit form for ease of administration and uniformity of dosage. Dosage unit form as used herein refers to physically discrete units suited as unitary dosages for the patient to be treated, each unit containing a pre-determined quantity of active compound or "effective amount" calculated to produce the desired therapeutic effect in association with the pharmaceutical carrier. The specification for the dosage unit forms of the invention are dictated by and directly dependent on the characteristics of the active compound, the particular therapeutic effect to be achieved, and the limitations inherent in the art of compounding such an active compound for the treatment of sensitivity in individuals.

[0033] The appropriate dosage or "effective amount" administered in any given case will, of course, vary depending upon known factors, such as the pharmacodynamic characteristics of the particular compound and its mode and route of administration; the age, general health, metabolism, weight of the patient and other factors which influence response to the compound; the nature and extent of the illness being treated; the kind of concurrent treatment, if any; the frequency of treatment; and the effect desired. Generally, the effective amount will be that amount of the present compounds needed to inhibit histone deacetylase, without resulting in toxicity to the patient. Inhibition at any level is within the scope of the present invention and will contribute to a therapeutic benefit in a patient. A daily dosage of active ingredient will typically be between about 10 to 10,000 milligrams per meter^2 (mg/m^2) of body mass, with the preferred dose being 50-5,000 mg/m^2 body mass.

[0034] It will be appreciated that the therapeutic benefits of administration of the present compounds will be manifest in a variety of ways, depending on the patient and the illness being treated. More than one therapeutic benefit may be observed. The elicitation of any therapeutic benefit by the present methods is within the scope of the invention. "Treating" or "treatment" refers herein to both therapeutic and prophylactic treatments; for ease of reference, "therapeutic

benefit" therefore refers collectively to both therapeutic and prophylactic benefits. Therapeutic benefits that may be achieved according to the present invention include, for example, retarding or eliminating tumor growth, apoptosis of tumor cells, healing wounds, healing cutaneous ulcers, ameliorating gastrointestinal disorders, modulating gene expression, inhibiting telomerase activity, inducing tolerance to antigens, preventing and/or ameliorating protozoan infection, inhibiting histone deacetylace in cells, modulating an immune response, ameliorating the effects of a cytotoxic agent, stimulating hematopoietic cells *ex vivo* and protecting hair follicles.

[0035] Modulation of an immune response can include, for example, enhancing cytokine secretion, inhibiting or delaying apoptosis in polymorphonuclear cells, enhancing polymorphonuclear cell function by augmenting hematopoietic growth factor secretion, inducing expression of cell surface antigens in tumor cells, and enhancing progenitor cell recovery after bone marrow transplantation.

[0036] Ameliorating the effects of a cytotoxic agent involves administering the present compounds in conjunction with the cytotoxic agent in such an amount so as to induce growth arrest of rapidly-proliferating epithelial cells of the patient, thereby protecting them from the cytotoxic effects of the agent. Cytotoxic agents, include, for example, chemotherapeutic agents, anticancer agents, and radiation therapy.

[0037] Modulating gene expression can be used to enhance, augment or repress the expression of a gene of interest. When expression of the gene of interest is to be enhanced or augmented, the gene can encode a gene product that is or acts as a repressor of another gene, a tumor suppressor, an inducer of apoptosis or an inducer of differentiation. Enhancing recombinant gene expression can be effected in a number of cells; the gene product can be any protein or peptide of interest such as tumor suppression genes. When expression of the gene of interest is to be repressed, the gene can encode a gene product that is or acts as an oncogene or an inhibitor of apoptosis, such as the bcl2 gene.

[0038] Inhibition of telomerase activity in cancer cells inhibits the malignant progression of the cells.

[0039] Inducing tolerance to an antigen is preferably carried about with a self-antigen, such as those associated with an autoimmune disease such as systemic lupus erythromatosus, rheumatoid arthritis, multiple sclerosis or diabetes. Tolerance can also be induced to one or more antigens present on a transplanted organ or cells.

[0040] The present invention is also directed a pharmaceutical composition comprising the compounds of formula 1 within a pharmaceutically acceptable carrier.

EXAMPLES

[0041] The following examples are intended to illustrate the invention, and should not be construed as limiting the invention in any way.

Example 1

Preparation of phenylpropiolioyloxymethyl acetate 3

[0042] Reference numerals correspond with those of Figure 1.

[0043] Phenylpropiolic acid (10 g) **1** and chloromethyl acetate (7.40 g) **2** were dissolved in 30 mL of DMF; 11.4 mL of TEA was added dropwise. The reaction was stirred overnight at room temperature

[0044] The next day GC analysis of reaction mixture showed the reaction was complete. (9.98 min; 98.6% A) The mixture was filtered and the precipitate was washed with 25 mL EtOAc. EtOAc (25 mL) was added to the filtrate, and the filtrate was washed with 50 mL of DI H_2O and 25 mL portions each of the following: HCl (5% wt.), $NaHCO_3$ (sat), and NaCL (sat). The organic phase was dried over Na_2SO_4, filtered, and evaporated. NMR of crude product wt. 14.10. g.

[0045] The next day, Kugelrohr distillation (100-105°C at 50 mtorr) of the crude yellow-orange material yielded 13.66 g of a light yellow liquid. DMF was still present, so the material was placed under hivac at 50°C for an additional 30 minutes. The final wt. of the product **3** was 13.50 g.

[0046] Table 1 provides a summary of components used.

TABLE 1

Compound	MW	moles	grams	mL	Density
phenylpropiolic acid **1**	146.15	0.07	10.0	--	--
chloromethyl acetate **2**	108.52	0.07	7.4	--	--
TEA	101	0.08	8.3	11.4	0.726
DMF	73.09	--	--	26.1	0.940

Example 2

Preparation of S-benzylthioglycoloyloxymethyl acetate **5**

[0047] Reference numerals correspond with those of Figure 2.

[0048] A solution of 10 g of S-benzylthioglycolic acid **4** and 6 g of chloromethyl acetate **2** in 50 mL of DMF was treated with 9.2 mL of TEA; the resulting solution was stirred overnight at room temperature.

[0049] The next day the reaction mixture was filtered to remove TEA·HCl and the filter cake was washed first with 50 mL of ethyl acetate then washed twice with water (50 mL), 5% hydrochloric acid (50 mL), sodium bicarbonate (50 mL) and brine (50 mL). The ethyl acetate layer was dried over sodium sulfate and concentrated in vacuo to afford 11.4 g of crude product. The proton NMR was perfect for the desired product **5** and GC indicated a high degree of purity.

[0050] The following day the product was distilled in a Kugelrohr at 130-135°C and 0.05 Torr to afford 11.2 g (80%) of product. The proton and carbon were perfect for the desired product **5**. GC indicated 100% purity.

[0051] Table 2 provides a summary of components used.

TABLE 2

Compound	MW	moles	grams	mL	Density
S-benzylthioglycolic acid **4**	182.24	0.05	10.0	--	--
chloromethyl acetate **2**	108.52	0.05	6.0	--	--
TEA	101	0.07	6.7	9.2	0.726
DMF	--	--	--	50	--

Example 3

Preparation of 3-(phenylthio)acryloyloxymethyl acetate **7**

[0052] Reference numerals correspond with those of Figure 3.

[0053] 3-(phenylthio)acrylic acid (10 g) **6** and chloromethyl acetate (6.0 g) **2** were dissolved in 25 mL of DMF; 9.3 mL of TEA was added dropwise. The reaction was stirred overnight at room temperature.

[0054] The next day, GC analysis of the reaction mixture showed a mixture of two products (11.33 min, 63.5% A; 10.82 min., 35.2% A). A GC trace of the starting acid showed a 1:2 mixture of compounds. The mixture was filtered and the precipitate was washed with 25 mL EtOAc. EtOAc (25 mL) was added to the filtrate, and the filtrate was washed with 50 mL of DI H_2O, and 25 mL portions each of the following: HCl (5% wt.), $NaHCO_3$ (sat), and NaCL (sat). The organic phase was dried over Na_2SO_4, filtered, and evaporated. The NMR crude product wt. was 13.64 g.

[0055] The next day Kugelrohr distillation (125-130°C at 50 mtorr) of the crude yellow-orange material **7** yielded 12.91 g of a light yellow liquid.

[0056] Table 3 provides a summary of compounds used.

TABLE 3

Compound	MW	moles	grams	mL	Density
3-(phenylthio)acrylic acid **6**	180.23	0.06	10.0	--	--
chloromethyl acetate **2**	108.52	0.06	6.0	--	--
TEA	101	0.07	6.7	9.3	0.726
DMF	73.09	--	--	21.2	0.940

Example 4

Preparation of 4-cyclohexylbutanoyloxymethyl acetate **9**

[0057] Reference numerals correspond with those of Figure 4.

[0058] 4-cyclohexanebutyric acid (10.0 g) **8** and chloromethyl acetate (6.4 g) **2** were dissolved in 25 mL of DMF; TEA (10.0 mL) was added dropwise over 10 minutes. The reaction mixture was stirred overnight at room temperature.

[0059] The next day GC analysis indicated the reaction was approximately two-thirds complete (acid 7.60 min.,

33.9% A; oxymethyl acetate 9.36 min., 66.1% A). The reaction was stirred overnight.

[0060] The following day GC analysis indicated a complete reaction (acid 7.71 min., 6.26% A; oxymethyl acetate 9.51 min., 90.48% A). The mixture was filtered; the precipitate was washed with 25 mL of EtOAc. 50 mL of EtOAc was added to the filtrate and the organic phase was washed with 1x50 mL of DI H_2O. The organic phase was then washed with a single 25 mL portion of each of the following: 5% wt. HCl, $NaHCO_3$ (sat), and NaCl (sat), then dried over Na_2SO_4 (anhy). Evaporation of solvent afforded 12.42 g of a clear liquid. NMR showed DMF. Kugelrohr distillation at 90-95°C and 65 mtorr yielded 10.35 g of a clear liquid **9**. NMR; GC; C13 NMR pkg. wt. 10.06 g.

[0061] Table 4 provides a summary of compounds used.

TABLE 4

Compound	MW	moles	grams	mL	Density
4-cyclohexanebutyric acid **8**	170.25	0.06	10.0	--	--
chloromethyl acetate **2**	108.52	0.06	6.4	--	--
TEA	101	0.07	7.1	9.8	0.726
DMF	73.09	--	--	22.4	0.940

Example 5

Preparation of 6-phenyl-3,5-hexadienoyloxymethyl acetate **11**

[0062] Reference numerals correspond to those in Figure 5.

[0063] A solution of 7 g of 6-phenyl-3,5-hexadienoic acid 10 and 4 g of chloromethyl acetate **2** was dissolved in 25 mL of DMF; 6.2 mL of TEA was added dropwise over 20 minutes. The resulting solution was stirred at room temperature and monitored by TLC. The reaction was stirred overnight at room temperature.

[0064] The next day, after 20 hours at room temperature, the total precipitated TEA-HCl was 4.8 g versus 5.5 g theoretical. The reaction was stirred for another four hours and there was no further precipitate. The reaction mixture was partitioned between 100 mL of ethyl acetate and 50 mL of water. The aqueous layer was acidified and extracted with ethyl acetate, dried over sodium sulfate and concentrated in vacuo to afford 0.6 g of the starting acid. The ethyl acetate extracts from above were washed with 50 mL each of 5% hydrochloric acid, saturated sodium bicarbonate and brine, dried over sodium sulfate and concentrated in vacuo to afford 7.7 g of a red orange oil **11**. The TLC (1:1 hexane: ethyl acetate) showed two distinct isomers except that the Rfs were very similar. The NMR showed that three isomers are present but that the E,E isomer predominates.

[0065] The following day the oil was dissolved in 20 mL of ethyl acetate and hexanes were added until it was just turbid (20 mL). Enough EtOAc (< 1 mL) was added to get the solution clear and then it was charged onto a Biotage 75S column.

[0066] The solution was eluted with 90:10 hexane ethyl acetate. Because of the need to use 1:1 ethyl acetate to solubilize the material, elution began in the second fraction. Better separation of the isomers might be possible if not for the insolubility in the elution solution.

[0067] Fraction 2-3 by TLC appeared to contain almost equal amounts of the E,E- and E,Z-isomer while a third unidentified isomer was also present. This fraction weighed less than 0.1 g and was discarded.

[0068] Fractions 4-11 were a mixture of three isomers with the E,E-isomer accounting for around 75-80% of the total. The E,Z-isomer was too small to even integrate although it is still present. The proton and carbon NMRs were perfect for the above product. The yield of these fractions was 3.5 g.

[0069] Fraction 12 appeared by TLC to be the E,E isomer but upon concentration and NMR analysis it was found to contain about 15% E,Z. The yield was about 0.3 g.

[0070] Fraction 13 was mostly the E,E-isomer although a trace of E,Z- could be seen by NRM. The carbon NMR was perfect for the desired E,E-isomer but there was less than 0.1 g. Fractions 12 and 13 were combined.

[0071] Table 5 provides a summary of components used.

TABLE 5

Compound	MW	moles	grams	mL	Density
6-phenyl-3,5-dienoic acid **10**	188.22	0.04	7.0	--	--
chloromethyl acetate **2**	108.52	0.04	4.0	--	--

TABLE 5 (continued)

Compound	MW	moles	grams	mL	Density
TEA	101	0.04	4.5	6.2	0.726
DMF	--	--	--	25	--

Example 6

Preparation of 5-phenyl-2,4-pentadienoyloxymethyl acetate **13**

[0072] Reference numerals correspond to those in Figure 6.

[0073] 5-phenylpentadienoic acid (7.5 g) **12** and chloromethyl acetate (4.3 g) **2** were dissolved in 25 mL of DMF; TEA (6.7 mL) was added dropwise over 10 minutes. The reaction mixture was stirred overnight at room temperature.

[0074] The next day the precipitate that formed overnight was collected by filtration and washed with 25 mL of ethyl acetate. The filtrate was partitioned between 50 mL each of water and ethyl acetate. The ethyl acetate layer was washed with 25 mL each of 5% hydrochloric acid, saturated sodium bicarbonate and brine, dried over sodium sulfate and concentrated in vacuo to afford 8.1 g of a liquid which by NMR was largely the desired product **13**. GC and TLC analysis showed very minor impurities (> 95% pure).

[0075] The following day the crude product was taken up in 10 mL of chloroform and loaded onto a Biotage 40M column and eluted with chloroform. Fractions 7-29 were combined and concentrated in vacuo to afford 5.8 g of a mobile liquid. The proton and carbon were perfect for the desired product **13**.

[0076] Table 6 provides a summary of components used.

TABLE 6

Compound	MW	moles	grams	mL	Density
5-phenylpentadienoic acid **12**	188.22	0.04	7.5	--	--
chloromethyl acetate **2**	108.52	0.04	4.3	--	--
TEA	101	0.05	4.8	6.7	0.726
DMF	--	--	--	25	--

Example 7

Preparation of cinnamoyloxymethyl acetate **15**

[0077] Reference numerals correspond with those of Figures 6 and 7.

[0078] Cinnamic acid (10 g) **14** and chlormethyl acetate (7.3 g) **2** were dissolved in 25 mL of DMF; 11.3 mL of TEA was added dropwise. The reaction was stirred overnight at room temperature.

[0079] A precipitate formed overnight. The reaction mixture was analyzed by GC and only a single signal was present. The reaction mixture was filtered and the filter cake was washed with 25 mL of ethyl acetate. The filtrate was partitioned between 50 mL each of water and ethyl acetate. The ethyl acetate layer was washed with 25 mL each of 5% hydrochloric acid, saturated sodium bicarbonate and brine, dried over sodium sulfate and concentrated in vacuo to afford a liquid which by NMR was largely the desired product **15**.

[0080] The next day the reaction mixture was distilled via Kugelrohr at 90-100°C and 0.2 Torr to afford 11.9 g of a clear liquid. The proton and carbon were perfect for the desired product **15**. The GC indicated 100% purity.

[0081] Table 7 provides a summary of components used.

TABLE 7

Compound	MW	moles	grams	mL	Density
cinnamic acid **14**	148.16	0.07	10.0	--	--
chloromethyl acetate **2**	108.52	0.07	7.3	--	--
TEA	101	0.08	8.2	11.3	0.726
DMF	--	--	--	25.8	--

Example 8

[0082] Compounds prepared according to Examples 1-7 were tested for anti-proliferation against PC-3 prostate breast cancer cells. Butyric acid and trichostatin, a potent HDAC inhibitor, were used as reference compounds. Both compounds were purchased from Sigma-Aldrich, Milwaukee, Wisconsin.

[0083] The PC3 cell line was maintained in RPMI supplemented with 10% fetal calf serum and antibiotics. The EDR assay was performed as described by Kern and Weisenthal in "Highly Specific Prediction of Antineoplastic Drug Resistance With An *In Vitro* Assay Using Suprapharmacologic Drug Exposures," *J. Nat. Cancer Inst.*, 82:582-588 (1990); and Fruehauf and Bosanquet, *"In vitro* Determination of Drug Response: A Discussion of Clinical Applications," *PPO Updates* 7(12):1-16 (1993). Cells were suspended in 0.12% soft agar in complete medium and plated (2,000 cells per well, as determined by preliminary experiments) in different drug concentrations onto a 0.4% agarose underlayer in 24-well plates. Plating cells on agarose underlayers supports the proliferation only of the transformed cells, ensuring that the growth signal stems from the malignant component of the tumor.

[0084] All compounds were dissolved in DMSO to 200x stock solutions. Stock solutions were diluted to 20x working solutions using the tissue culture medium, serially diluted and added to the 24-well plates. The concentration range was 0.001 μM to 0.3 μM for trichostatin and 10 μM - 1,000 μM for the other compounds. No significant changes in pH of the culture medium were observed under the above conditions. Diluent control wells contained PC3 cells treated with DMSO, at the dilutions used for appropriate drug treatment. All experimental points were represented by two separate wells (duplicates). Positive controls were determined using at least two wells treated with an extremely high dose of cisplatin, an anti-cancer agent. Four wells containing tumor cells that were not treated with drugs served as negative controls in each experiment.

[0085] Cells were incubated with drugs under standard culture conditions for five days. Cultures were pulsed with tritiated thymidine (^{3}H-TdR, New Life Science Products, Boston, MA) at 5 μCi per well for the last 48 hours of the culture period. Cell culture plates were then heated to 90°C to liquefy the agarose, and cells were harvested onto glass fiber filters, which were then placed into counting vials containing liquid scintillation fluid. The radioactivity trapped on the filters was counted with a Beckman scintillation counter. The fraction of surviving cells was determined by comparing ^{3}H-TdR incorporation in treated (experimental points) and untreated (negative control) wells. All drug concentrations are presented as μM, allowing for normalization of drug response curves and direct comparison of the effects of the drugs. Microsoft Excel was used to organize the raw data on EDR experiments, and the SigmaPlot program was utilized to generate drug response curves. All drug response curves were as approximated as sigmoidal equations (characteristic for typical drug response curves) to fit the data. IC_{50} values were determined using the approximated sigmoidal curves and expressed as μM.

[0086] Table 8 provides the PC-3 IC_{50} data for each of the compounds tested.

TABLE 8

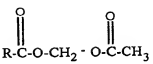
COMPOUND	PC-3 IC_{50} (μM)
phenylpropioloyloxymethyl acetate	25
S-benzylthioglycoloyloxymethyl acetate	28.4
3-(phenylthio)acryloyloxymethyl acetate	30
4-cyclohexanebutyroyloxymethyl acetate	23.2
6-phenyl-3,5-hexadienoyloxymethyl acetate	18
5 -phenyl-2,4-pentadienoyloxymethyl acetate	20
cinnamoyloxymethyl acetate	12.5
butyric acid	> 2000
trichostatin	0.005

[0087] As can be seen from Table 8, the results demonstrate that the compounds of the present invention possess superior activity as compared to butyric acid.

[0088] Whereas particular embodiments of this invention have been described above for purposes of illustration, it will be evident to those skilled in the art that numerous variations of the details of the present invention may be made without departing from the invention as defined in the appended claims.

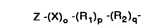
Claims

1. A compound having the formula (1):



(1)

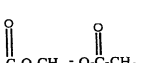
wherein R is substituted or unsubstituted C_2-C_{10} alkenyl, substituted or unsubstituted C_2-C_{10} alkynyl, a cis or trans retinoyl group, or has the general formula (2):



(2)

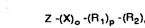
wherein Z is selected from the group consisting of hydrogen and substituted or unsubstituted aryl, heteroaryl, cycloalkyl having the formula C_nH_{2n-1}, and alkoxy;
wherein n is 3 or greater;
wherein X is S, O, C=O or CH_2;
wherein R_1 is S, O, CH=CH or C≡C;
wherein R_2 is CH_2, CH=CH or C≡C; and
wherein o, p and q are the same or different are each between 0 and 10, but when o is zero and R_1 or R_2 is CH=CH or C≡C, Z is not hydrogen or alkoxy.

2. The compound of Claim 1, wherein R is C_6H_5-C≡C-.
3. The compound of Claim 1, wherein R is C_6H_5-CH_2-S-CH_2-.
4. The compound of Claim 1, wherein R is C_6H_5-S-CH=CH-.
5. The compound of Claim 1, wherein R is C_6H_{11}-$(CH_2)_3$-.
6. The compound of Claim 1, wherein R is C_6H_5-CH=CH-.
7. The compound of Claim 1, wherein R is C_6H_5-$(CH=CH)_2$-CH_2-.
8. The compound of Claim 1, wherein R is C_6H_5-$(CH=CH)_2$-.
9. Use of a compound having the formula (1):



(1)

wherein R is substituted or unsubstituted C_2-C_{10} alkenyl, substituted or unsubstituted C_2-C_{10} alkynyl, a cis or trans retinoyl group, or has the general formula (2):



(2)

wherein Z is selected from the group consisting of hydrogen and substituted or unsubstituted aryl, heteroaryl, cycloalkyl having the formula C_nH_{2n-1}, and alkoxy;

wherein n is 3 or greater;

wherein X is S, O, C=O or CH_2;

wherein R_1 is S, O, CH=CH or C≡C;

wherein R_2 is CH_2, CH=CH or C≡C; and

wherein o, p and q are the same or different are each between 0 and 10, but when o is zero and R_1 or R_2 is CH=CH or C≡C, Z is not hydrogen or alkoxy for the manufacture of a medicament for treating an illness in a patient.

10. The use of Claim 9 wherein said illness is cancer, hemological disease or an inherited metabolic disease.

11. The use of Claim 9 wherein said medicament is administered at an amount of about 10 milligrams per meter^2 of body mass, per day.

12. The use of Claim 9 wherein said medicament is administered orally, parenterally, transdermally, transmucosally, intranasally, rectally or topically.

13. The use of Claim 9 wherein said treatment results in one or more therapeutic benefits selected from the group consisting of retarding or eliminating tumor growth, inducing apoptosis of tumor cells, healing wounds, healing cutaneous ulcers, ameliorating gastrointestinal disorders, modulating gene expression, inhibiting telomerase activity, inducing tolerance to antigens, preventing or ameliorating protozoan infection, inhibiting histone deacetylace in cells, modulating an immune response, ameliorating the effects of a cytotoxic agent, stimulating hematopoietic cells *ex vivo* and protecting against injury to hair follicles.

14. A pharmaceutical composition comprising an effective amount of a compound of Claim 1 and a pharmaceutically acceptable carrier.

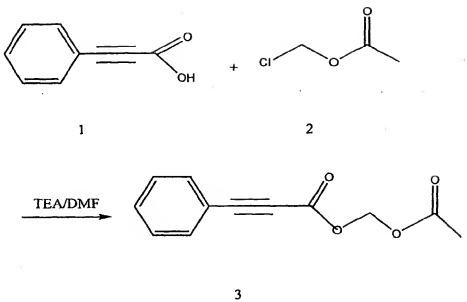


Figure 1

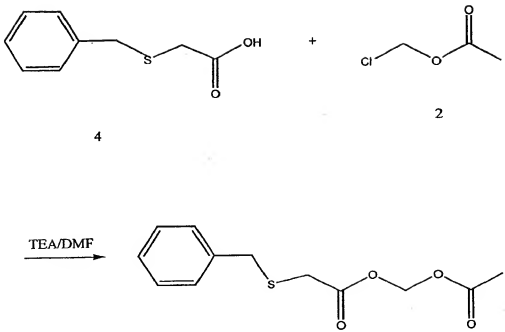


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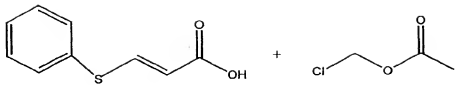
Figure 2



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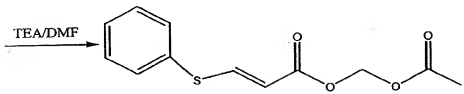

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Figure 3

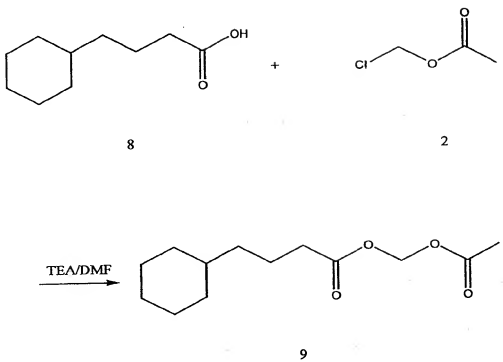
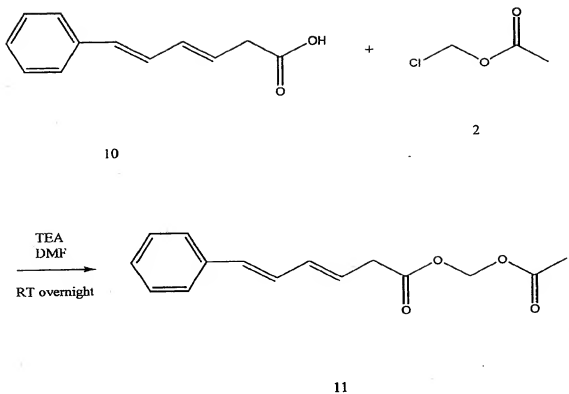


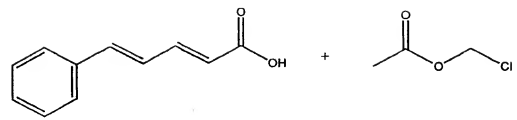
Figure 4



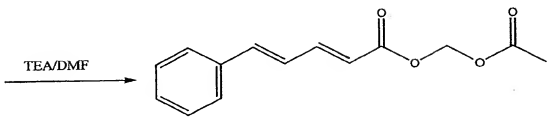
10 2

11

Figure 5

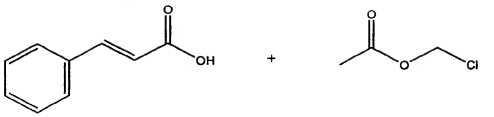


12 2



13

Figure 6



14 2

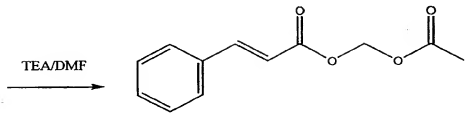


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Figure 7

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PARTIAL EUROPEAN SEARCH REPORT

Application Number EP 01 31 0692

which under Rule 45 of the European Patent Convention shall be considered, for the purposes of subsequent proceedings, as the European search report

DOCUMENTS CONSIDERED TO BE RELEVANT

Category	Citation of document with indication, where appropriate, of relevant passages	Relevant to claim	CLASSIFICATION OF THE APPLICATION (Int.Cl.7)
X	GB 1 138 745 A (BEECHAM GROUP LTD) 1 January 1969 (1969-01-01) * claim 1 *	1,9,12, 14	C07C69/608 C07C69/612 C07C323/52 A61K31/215 A61P35/00
X	GB 1 579 531 A (GLAXO LAB LTD) 19 November 1980 (1980-11-19) * example 109 *	1,9,12, 14	
X	GB 2 043 639 A (ERBA FARMITALIA) 8 October 1980 (1980-10-08) * examples 1-7,9-16,18,19,26-29,35-44,47-51 *	1,9,12, 14	
X	GB 2 048 857 A (ERBA FARMITALIA) 17 December 1980 (1980-12-17) * examples 27,28 *	1,9,12, 14	
X	GB 2 097 786 A (ERBA FARMITALIA) 10 November 1982 (1982-11-10) * examples 1A,11A,15A,16A *	1,9,12, 14	TECHNICAL FIELDS SEARCHED (Int.Cl.7)
X	JP 57 179190 A (YAMANOUCHI SEIYAKU KK) 4 November 1982 (1982-11-04) * examples 6-9 *	1,9,12, 14	C07C A61K
	-/--		

INCOMPLETE SEARCH

The Search Division considers that the present application, or one or more of its claims, does/do not comply with the EPC to such an extent that a meaningful search into the state of the art cannot be carried out, or can only be carried out partially, for these claims.

Claims searched completely :

Claims searched incompletely :

Claims not searched :

Reason for the limitation of the search:

see sheet C

Place of search	Date of completion of the search	Examiner
MUNICH	12 March 2002	Janus, S

CATEGORY OF CITED DOCUMENTS

X : particularly relevant if taken alone
Y : particularly relevant if combined with another document of the same category
A : technological background
O : non-written disclosure
P : intermediate document

T : theory or principle underlying the invention
E : earlier patent document, but published on, or after the filing date
D : document cited in the application
L : document cited for other reasons

& : member of the same patent family, corresponding document

EPO FORM 1503 03.82 (P04C01)

European Patent Office

INCOMPLETE SEARCH SHEET C

Application Number
EP 01 31 0692

Claim(s) searched completely:
2-8

Claim(s) searched incompletely:
1, 9-14

Reason for the limitation of the search:

The initial phase of the search revealed a very large number of chemical compounds (894) potentially relevant to the issue of novelty. So many documents were then retrieved (987) that it is impossible to determine which parts of the claim(s) may be said to define subject-matter for which protection might legitimately be sought (Article 84 EPC). For these reasons, a meaningful search over the whole breadth of the claim(s) is impossible.

In addition, the present claim 1 relate to an extremely large number of possible compounds. Support within the meaning of Article 84 EPC is to be found, however, for only a very small proportion of the compounds claimed. In the present case, the claims so lack support that a meaningful search over the whole of the claimed scope is impossible. Consequently, the search has been carried out for those parts of the claims which appear to be supported and disclosed, namely those parts relating to the compounds of formula (I) wherein R includes a phenyl or a cyclohexyl group linked to the -C(=O)-O-CH2-O-C(=O)-CH3 group by a chain comprising between 2 and 5 non-hydrogen atoms

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PARTIAL EUROPEAN SEARCH REPORT

Application Number EP 01 31 0692

DOCUMENTS CONSIDERED TO BE RELEVANT			CLASSIFICATION OF THE APPLICATION (Int.Cl.7)
Category	Citation of document with indication, where appropriate, of relevant passages	Relevant to claim	
X	DE 32 07 241 A (SCHERING AG) 1 September 1983 (1983-09-01) * claims 32,35 *	1,9,12, 14	
X	JP 60 139682 A (MITSUI TOATSU KAGAKU KK) 24 July 1985 (1985-07-24) * claim 2 *	1,9,12, 14	
E	WO 01 90136 A (SAREK JAN ;HAJDUCH MARIAN (CZ); UNIV KARLOVA (CZ); UNIVERZITA PALA) 29 November 2001 (2001-11-29) * examples I.19,I.21,I.28,I.41 *	1,9-14	
X	LOMBARDI P ET AL: "TOTAL SYNTHESIS OF THIA ANALOGUES OF CLAVULANIC ACID" TETRAHEDRON LETTERS, ELSEVIER SCIENCE PUBLISHERS, AMSTERDAM, NL, no. 39, 1979, pages 3777-3780, XP001057691 ISSN: 0040-4039 * compounds 8-10 and 13-16 wherein R3 is CH2OCOCH3 *	1	TECHNICAL FIELDS SEARCHED (Int.Cl.7)
X	FOGLIO P ET AL: "Synthesis of Optically Active (5R)-2-Penem-3-carboxylates from penicillanic Acid Derivatives : New Potent Anti-bacterial Agents" J.C.S. CHEM. COMMUN., no. 2, 1980, pages 70-71, XP001057729 London * compounds 1c, 1d, 5b, 6b, 10 *	1	
	-/--		

EPO FORM 1503 03.82 (P04C01)

European Patent Office

PARTIAL EUROPEAN SEARCH REPORT

Application Number
EP 01 31 0692

DOCUMENTS CONSIDERED TO BE RELEVANT			CLASSIFICATION OF THE APPLICATION (Int.Cl.7)
Category	Citation of document with indication, where appropriate, of relevant passages	Relevant to claim	
X	DREW J H ET AL: "ANTIFERTILITY ACTIVITY OF N-PROTECTED GLYCINE ACTIVATED ESTERS" JOURNAL OF PHARMACEUTICAL SCIENCES, AMERICAN PHARMACEUTICAL ASSOCIATION. WASHINGTON, US, vol. 70, no. 1, January 1981 (1981-01), pages 60-63, XP001057684 ISSN: 0022-3549 * compound XI *	1	
X	LONGO A ET AL: "Total Synthesis and Antibacterial Activity of Some 6-Ethyl-2-penem Derivatives" GAZZ. CHIM. ITAL., vol. 111, no. 9-10, 1981, pages 371-377, XP001057735 * compounds 15-24 *	1	TECHNICAL FIELDS SEARCHED (Int.Cl.7)
X	ALPEGIANI M ET AL: "SYNTHESIS OF 2-(HETEROCYCLYLTHIOMETHYL)PENEMS" GAZZETTA CHIMICA ITALIANA, SOCIETA CHIMICA ITALIANA, ROME, IT, vol. 114, no. 7/8, 1984, pages 319-324, XP001057659 ISSN: 0016-5603 * compounds 1b, 2b, 3b, 13b *	1	
X	BATTISTINI C ET AL: "Synthesis of New Penem Derivatives : N-Substituted Analogs of FCE 22101" HETEROCYCLES, vol. 23, no. 8, 1985, pages 1929-1932, XP001057252 * compound 1b *	1	

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EPO FORM 1503 03.82 (P04C10)

European Patent Office

PARTIAL EUROPEAN SEARCH REPORT

Application Number
EP 01 31 0692

DOCUMENTS CONSIDERED TO BE RELEVANT			CLASSIFICATION OF THE APPLICATION (Int.Cl.7)
Category	Citation of document with indication, where appropriate, of relevant passages	Relevant to claim	
X	HEILPORN S ET AL: "Synthesis of BAPTA-AM Analogues Capable of Enhancing the Vascular Production of Prostacyclin " BULL. SOC. CHIM. BELG., vol. 103, no. 7-8, 1994, pages 309-319, XP001057725 * compounds 2, 15-17, 26, 35, 41, 46, 47 *	1	
X	SALAZAR L ET AL: "Optically-active Dihydropyridines via Lipase-catalyzed Enantioselective Hydrolysis" TETRAHEDRON: ASYMMETRY, ELSEVIER SCIENCE PUBLISHERS, AMSTERDAM, NL, vol. 6, no. 12, 1 December 1995 (1995-12-01), pages 2917-2920, XP004047939 ISSN: 0957-4166 * page 2918, Scheme *	1	TECHNICAL FIELDS SEARCHED (Int.Cl.7)
Y	EP 0 302 349 A (UNIV BAR ILAN ;KUPAT HOLIM HEALTH INSURANCE (IL)) 8 February 1989 (1989-02-08) * formula (I) *	1-14	
Y	WO 98 00127 A (UNIV BAR ILAN ;KUPAT HOLIM HEALTH INSURANCE (IL); REPHAELI ADA (US) 8 January 1998 (1998-01-08) * abstract *	1-14	
Y	WO 98 39965 A (BEACON LAB L L C) 17 September 1998 (1998-09-17) * abstract; claim 1 *	1-14	
Y	WO 98 39966 A (BEACON LAB L L C) 17 September 1998 (1998-09-17) * abstract; claim 1 *	1-14	

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Application Number
EP 01 31 0692

DOCUMENTS CONSIDERED TO BE RELEVANT			CLASSIFICATION OF THE APPLICATION (Int.Cl.7)
Category	Citation of document with indication, where appropriate, of relevant passages	Relevant to claim	
Y	WO 98 40065 A (BEACON LAB L L C) 17 September 1998 (1998-09-17) * abstract; claim 1 *	1-14	
Y	WO 98 40066 A (BEACON LAB L L C) 17 September 1998 (1998-09-17) * abstract; claim 1 *	1-14	
Y	WO 98 40078 A (BEACON LAB L L C) 17 September 1998 (1998-09-17) * abstract; claim 1 *	1-14	
			TECHNICAL FIELDS SEARCHED (Int.Cl.7)

EPO FORM 1503 03.82 (P04C10)

ANNEX TO THE EUROPEAN SEARCH REPORT ON EUROPEAN PATENT APPLICATION NO. EP 01 31 0692

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The European Patent Office is in no way liable for these particulars which are merely given for the purpose of information.

12-03-2002

Patent document cited in search report		Publication date	Patent family member(s)		Publication date
GB 1138745	A	01-01-1969	NONE		
GB 1579531	A	19-11-1980	AU	2270977 A	31-08-1978
			BE	851821 A1	25-08-1977
			DE	2708330 A1	08-09-1977
			DK	86277 A	28-08-1977
			FR	2342292 A1	23-09-1977
			JP	52125191 A	20-10-1977
			JP	61026547 B	20-06-1986
			NL	7702027 A	30-08-1977
			SE	7702139 A	10-10-1977
			ZA	7701140 A	25-01-1978
GB 2043639	A	08-10-1980	AT	368506 B	25-10-1982
			AT	91980 A	15-02-1982
			AU	535080 B2	01-03-1984
			AU	5567080 A	11-09-1980
			BE	881862 A1	22-08-1980
			CA	1154010 A1	20-09-1983
			CA	1212665 B	14-10-1986
			CH	651570 A5	30-09-1985
			CH	654831 A5	14-03-1986
			CS	226010 B2	19-03-1984
			CS	226027 B2	19-03-1984
			DE	3006273 A1	04-09-1980
			DK	77580 A ,B,	25-08-1980
			ES	488886 D0	16-11-1981
			ES	8200685 A1	01-02-1982
			ES	495977 D0	01-10-1981
			ES	8107224 A1	16-12-1981
			FI	800493 A ,B,	25-08-1980
			FR	2449690 A1	19-09-1980
			GB	2114132 A ,B	17-08-1983
			GB	2116167 A ,B	21-09-1983
			GB	2104893 A ,B	16-03-1983
			GR	73623 A1	26-03-1984
			HK	74487 A	23-10-1987
			HU	182664 B	28-02-1984
			IE	49407 B1	02-10-1985
			IE	49408 B1	02-10-1985
			IE	49409 B1	02-10-1985
			IE	49410 B1	02-10-1985
			IL	67089 A	31-05-1985
			IL	67090 A	31-05-1985
			IT	1193922 B	31-08-1988
			JP	1638842 C	31-01-1992

EPO FORM P0459

For more details about this annex : see Official Journal of the European Patent Office, No. 12/82

ANNEX TO THE EUROPEAN SEARCH REPORT ON EUROPEAN PATENT APPLICATION NO. EP 01 31 0692

This annex lists the patent family members relating to the patent documents cited in the above-mentioned European search report. The members are as contained in the European Patent Office EDP file on
The European Patent Office is in no way liable for these particulars which are merely given for the purpose of information.

12-03-2002

Patent document cited in search report	Publication date	Patent family member(s)	Publication date
GB 2043639 A		JP 3000396 B	07-01-1991
		JP 55136288 A	23-10-1980
		LU 82192 A1	06-06-1980
		MX 9203121 A1	01-07-1992
		NL 8001012 A ,B,	26-08-1980
		NO 800501 A ,B,	25-08-1980
		NZ 192949 A	16-03-1984
		PT 70849 A	01-03-1980
		SE 449489 B	04-05-1987
		SE 8001424 A	25-08-1980
		SU 1186086 A3	15-10-1985
		YU 46180 A1	28-02-1983
		ZA 8000993 A	25-02-1981
		IL 59430 A	31-05-1985
		NZ 203161 A	30-03-1984
		SI 8010461 A8	31-10-1996
GB 2048857 A	17-12-1980	AT 369371 B	27-12-1982
		AT 177480 A	15-05-1982
		AU 536673 B2	17-05-1984
		AU 5701580 A	09-10-1980
		BE 882621 A1	03-10-1980
		CA 1148143 A1	14-06-1983
		CH 645645 A5	15-10-1984
		CS 219273 B2	25-03-1983
		DE 3012975 A1	23-10-1980
		DK 144180 A	07-10-1980
		FI 801080 A	07-10-1980
		FR 2453162 A1	31-10-1980
		GR 66673 A1	08-04-1981
		HU 182118 B	28-12-1983
		IE 49656 B1	13-11-1985
		IL 59760 A	31-07-1983
		IT 1196407 B	16-11-1988
		JP 55139385 A	31-10-1980
		LU 82320 A1	02-07-1980
		NL 8001919 A	08-10-1980
		SE 8002600 A	07-10-1980
		SU 1152524 A3	23-04-1985
		US 4269771 A	26-05-1981
		YU 93880 A1	28-02-1983
		ZA 8002000 A	29-04-1981
GB 2097786 A	10-11-1982	US 4482565 A	13-11-1984
		BG 60539 B2	28-07-1995
		CS 9104154 A3	17-06-1992

EPO FORM P0459

For more details about this annex : see Official Journal of the European Patent Office, No. 12/82

ANNEX TO THE EUROPEAN SEARCH REPORT ON EUROPEAN PATENT APPLICATION NO. EP 01 31 0692

This annex lists the patent family members relating to the patent documents cited in the above-mentioned European search report. The members are as contained in the European Patent Office EDP file on
The European Patent Office is in no way liable for these particulars which are merely given for the purpose of information.

12-03-2002

Patent document cited in search report	Publication date	Patent family member(s)	Publication date
GB 2097786 A		HK 74587 A	23-10-1987
		SG 34487 G	24-07-1987
		US 4558042 A	10-12-1985
JP 57179190 A	04-11-1982	NONE	
DE 3207241 A	01-09-1983	DE 3207241 A1	01-09-1983
JP 60139682 A	24-07-1985	NONE	
WO 0190136 A	29-11-2001	GB 2362648 A	28-11-2001
		WO 0190136 A2	29-11-2001
EP 0302349 A	08-02-1989	IL 87072 A	18-08-1993
		AT 95164 T	15-10-1993
		CA 1327595 A1	08-03-1994
		DE 3884517 D1	04-11-1993
		DE 3884517 T2	10-02-1994
		EP 0302349 A2	08-02-1989
		ES 2045028 T3	16-01-1994
		HK 1005447 A1	08-01-1999
		JP 1139545 A	01-06-1989
		JP 2763894 B2	11-06-1998
		JP 3044009 B2	22-05-2000
		JP 10152436 A	09-06-1998
		JP 2847078 B2	13-01-1999
		JP 10152454 A	09-06-1998
		US 5200553 A	06-04-1993
WO 9800127 A	08-01-1998	US 6040342 A	21-03-2000
		AU 3588097 A	21-01-1998
		CA 2258593 A1	08-01-1998
		EP 0927033 A1	07-07-1999
		WO 9800127 A1	08-01-1998
		US 6043277 A	28-03-2000
WO 9839965 A	17-09-1998	US 6124495 A	26-09-2000
		AU 6549698 A	29-09-1998
		EP 0973388 A1	26-01-2000
		WO 9839965 A1	17-09-1998
WO 9839966 A	17-09-1998	US 6110970 A	29-08-2000
		AU 6550098 A	29-09-1998
		EP 0973389 A1	26-01-2000
		WO 9839966 A1	17-09-1998

EPO FORM P0459

For more details about this annex : see Official Journal of the European Patent Office, No. 12/82

ANNEX TO THE EUROPEAN SEARCH REPORT ON EUROPEAN PATENT APPLICATION NO. EP 01 31 0692

This annex lists the patent family members relating to the patent documents cited in the above-mentioned European search report. The members are as contained in the European Patent Office EDP file on
The European Patent Office is in no way liable for these particulars which are merely given for the purpose of information.

12-03-2002

Patent document cited in search report	Publication date	Patent family member(s)	Publication date
WO 9840065 A	17-09-1998	US 6043389 A	28-03-2000
		AU 728419 B2	11-01-2001
		AU 6550198 A	29-09-1998
		EP 0998278 A1	10-05-2000
		JP 2001514664 T	11-09-2001
		WO 9840065 A1	17-09-1998
		US 6239176 B1	29-05-2001
WO 9840066 A	17-09-1998	US 6110955 A	29-08-2000
		AU 6457998 A	29-09-1998
		EP 0986380 A1	22-03-2000
		WO 9840066 A1	17-09-1998
WO 9840078 A	17-09-1998	US 5939455 A	17-08-1999
		AU 6547898 A	29-09-1998
		WO 9840078 A1	17-09-1998

EPO FORM P0459

For more details about this annex : see Official Journal of the European Patent Office, No. 12/82